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Forum Paper

A strategy for successful integration of DNA-based methods in aquatic monitoring

Philippe Blancher[1], Estelle Lefrançois[2], Frédéric Rimet[3], Valentin Vasselon[3,4], Christine Argillier[5], Jens Arle[6], Pedro Beja[7], Pieter Boets[8], Jeanne Boughaba[9], Christian Chauvin[10], Michael Deacon[11], Willie Duncan[12], Gunilla Ejdung[13], Stefania Erba[14], Benoit Ferrari[15], Helmut Fischer[16], Bernd Hänfling[17], Michael Haldin[18], Daniel Hering[19], Nicolas Hette-Tronquart[20], Alice Hiley[21], Marko Järvinen[22], Benjamin Jeannot[23], Maria Kahlert[24], Martyn Kelly[25], Julia Kleinteich[16], Serdar Koyuncuoğlu[26], Sascha Krenek[16], Sidsel Langhein-Winther[11], Florian Leese[19], David Mann[27], Rémy Marcel[28], Stefania Marcheggiani[29], Kristian Meissner[22], Patricia Mergen[30], Olivier Monnier[20], Frank Narendja[31], Diane Neu[32], Veronica Onofre Pinto[33], Alina Pawlowska[34], Jan Pawlowski[35], Martin Petersen[11], Sandra Poikane[36], Didier Pont[37], Marie-Sophie Renevier[38], Steinar Sandoy[39], Jonas Svensson[18], Rosa Trobajo[40], Andrea Tünde Zagyva[41], Iakovos Tziortzis[42], Berry van der Hoorn[43], Marlen Ines Vasquez[44], Kerry Walsh[21], Alexander Weigand[45], Agnès Bouchez[3]

1 *Blancher Consultant, Lyon, France*
2 *ECO in'EAU, Montferrier sur Lez, France*
3 *INRAE, USMB, CARRTEL, Thonon-les-Bains, France*
4 *Scimabio Interface, Thonon-les-Bains, France*
5 *INRAE, RECOVER, Aix-en-Provence, France*
6 *German Environment Agency, Dessau-Roßlau, Germany*
7 *CIBIO/InBio, University of Porto, Porto, Portugal*
8 *Provincial Centre of Environmental Research, Ghent, Belgium*
9 *European Commission, Brussel, Belgium*
10 *INRAE, EABX, Cestas, France*
11 *Danish Environmental Protection Agency, Gram, Denmark*
12 *Scottish Environment Protection Agency, Stirling, United Kingdom*
13 *Swedish Environmental Protection Agency (SEPA), Lund, Sweden*
14 *Water Research Institute, CNR-IRSA, Brugherio, Italy*
15 *Swiss Centre for Applied Ecotoxicology, Lausanne, Switzerland*
16 *German Federal Institute of Hydrology, BfG, Koblenz, Germany*
17 *University of Hull, Hull, United Kingdom*
18 *Swedish Agency for Marine and Water Management, Göteborg, Sweden*
19 *University of Duisburg-Essen, Duisburg-Essen, Essen, Germany*
20 *Office Français de la Biodiversité, OFB, Vincennes, France*
21 *UK Environment Agency, Rotherham, United Kingdom*
22 *Finnish Environment Institute, SYKE, Jyvaskyla, Helsinki, Finland*
23 *French Ministry for an Ecological Transition, MET, Paris, France*
24 *Swedish University of Agricultural Sciences, Department of Aquatic Sciences and Assessment, Uppsala, Sweden*
25 *Bowburn Consultancy, Bowburn, United Kingdom*
26 *Water Management, MoAF, Clermont-Ferrand, Ankara, Turkey*
27 *Royal Botanic Garden Edinburgh, Edinburgh, United Kingdom*
28 *Aquabio, Clermont-Ferrand, Clermont-Ferrand, France*
29 *Italian National Institute of Health, Roma, Italy*
30 *Meise Botanic Garden, Meise, Belgium*
31 *Environment Agency, Vienna, Austria*
32 *Administration de la gestion de l'eau, AGE, Luxembourg, Luxembourg*
33 *Portuguese Environment Agency, Amadora, Portugal*

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PENSOFT

34 *ID-Gene ecodiagnostics, Plan-les-Ouates, Switzerland*
35 *University of Geneva, Geneva, Switzerland*
36 *European Commission, Joint Research Centre (JRC), Ispra, Italy*
37 *BOKU-IHG, Vienna, Austria*
38 *Federal Office for the Environment, FOEN, Switzerland*
39 *Norwegian Environment Agency, Trondheim, Norway*
40 *Institute for Food and Agricultural Research and Technology, IRTA, Sant Carles de la Rapita, Catalonia, Spain*
41 *Department for Water Management and Water, OVF, Hungary*
42 *Water Development Department, Ministry of Agriculture, Rural Development and Environment, Cyprus*
43 *Inholland University of Applied Sciences, Delft, Leiden, Netherlands*
44 *Cyprus University of Technology, CUT, Limassol, Cyprus*
45 *National Museum of Natural History Luxembourg, MNHNL, Luxembourg, Luxembourg*

Corresponding author: Agnès Bouchez (agnes.bouchez@inrae.fr)

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Abstract

Recent advances in molecular biomonitoring open new horizons for aquatic ecosystem assessment. Rapid and cost-effective methods based on organismal DNA or environmental DNA (eDNA) now offer the opportunity to produce inventories of indicator taxa that can subsequently be used to assess biodiversity and ecological quality. However, the integration of these new DNA-based methods into current monitoring practices is not straightforward, and will require coordinated actions in the coming years at national and international levels.

To plan and stimulate such an integration, the European network DNAqua-Net (COST Action CA15219) brought together international experts from academia, as well as key environmental biomonitoring stakeholders from different European countries. Together, this transdisciplinary consortium developed a roadmap for implementing DNA-based methods with a focus on inland waters assessed by the EU Water Framework Directive (2000/60/EC). This was done through a series of online workshops held in April 2020, which included fifty participants, followed by extensive synthesis work.

The roadmap is organised around six objectives: 1) to highlight the effectiveness and benefits of DNA-based methods, 2) develop an adaptive approach for the implementation of new methods, 3) provide guidelines and standards for best practice, 4) engage stakeholders and ensure effective knowledge transfer, 5) support the environmental biomonitoring sector to achieve the required changes, 6) steer the process and harmonise efforts at the European level.

This paper provides an overview of the forum discussions and the common European views that have emerged from them, while reflecting the diversity of situations in different countries. It highlights important actions required for a successful implementation of DNA-based biomonitoring of aquatic ecosystems by 2030.

Key Words

Aquatic ecosystems, DNAqua-Net, Ecological assessment, eDNA, Water Framework Directive

Introduction

Global and local anthropogenic changes exert severe pressures on aquatic ecosystems, endangering their ecological status, biodiversity, and the services they provide. Monitoring practices and the European Union (EU) policies are seeking to address these key environmental and societal challenges and strive to implement governance models to manage aquatic ecosystems sustainably. Currently, the bioassessment of these ecosystems is based on various biotic metrics and indices, which mainly use taxonomic inventories, taxon abundance and diversity in selected groups of organisms to define the ecological status (Birk et al. 2012; Poikane et al. 2015; Charles et al. 2020). Overall, these conventional indices have proven their general suitability for a long time and have consequently been used for statutory biomonitoring. However, they have some limitations, often related to issues in the morphological identification of indicator taxa, their speed, and sometimes the invasive nature of their assessment. The cost also tends to be high, typically due to the morphological identification step, which is time-consuming, especially when assessments are based on species level identification, and requires highly skilled staff in taxonomy, who are increasingly rare.

Developments based on organismal DNA or environmental DNA (eDNA), can potentially alleviate some of these limitations, by using DNA sequences to identify organisms and characterise a given ecosystem. As proposed by Taberlet et al. (2012) and recently refined by Pawlowski et al. (2020a), here we use the term "eDNA" in the broadest sense, i.e. DNA isolated directly from any environmental sample (water, sediment etc.). DNA-based methods are making very swift progress and are opening up new horizons for assessing the biodiversity of aquatic environments (Hering et al. 2018; Vitecek et al. 2021). They offer the possibility to generate more detailed biodiversity inventories from environmental samples and improve taxonomic resolution in various groups of indicator organisms. All of this comes at a fraction of the cost and time of an equivalent conventional approach (Pawlowski et al 2018; Hyvärinen et al. 2021). For some bioindicators, e.g., fish fauna (Pont et al. 2021), the sampling procedure is also potentially simpler and less invasive. The inventories produced can then be used for a more accurate assessment of biodiversity and ecological quality (e.g., Apothéloz-Perret-Gentil et al 2021).

Faced with the rapid development of DNA-based methods and growing interest in their application in biodiversity research and biomonitoring, a European network DNAqua-Net (COST Action CA15219) was launched in 2016. Its aims were to advance the application of DNA-based methods and develop a roadmap to incorporate them in standardised ecological assessments of aquatic ecosystems in Europe and beyond (Leese et al. 2016, 2018). The implementation of these DNA-based methods in monitoring practices is not straightforward and involves significant technical and organisational changes. Furthermore, their introduction involves overcoming social barriers, political and governance obstacles, cultural resistance to change, and subsequently the standardisation and operationalisation of methods to make them accessible via fit-for-purpose infrastructure, ensure compatibility and cross-validation with current methods, and train a new generation of professionals. Thus, their adoption will require coordinated actions at national and international levels (Leese et al. 2018).

To develop a roadmap for implementing the novel methods, two science-policy workshop series have been organised in recent years. The first one was organised in 2019 in the framework of SYNAQUA (an INTERREG France-Switzerland research project led by INRAE and the University of Geneva; Lefrançois et al. 2018). Interactive breakout sessions brought together 46 French and Swiss public decision-makers and environmental management professionals from 34 organisations. Different possible scenarios for applying DNA-based methods in regulatory biomonitoring were identified and finally a realistic "virtuous scenario" was designed, with an action plan to facilitate its implementation (Bouchez et al. 2021). The second series was organised by DNAqua-Net (CA15219) and INRAE (France) in 2020 to broaden the perspective to EU level. The action plan developed during the SYNAQUA workshops served as a working basis, and DNAqua-Net experts and environmental stakeholders from different European countries were involved. The discussions and outcomes of the DNAqua-Net workshops were compiled in an open-access report, reflecting the diversity of situations in Europe, as well as common views (Lefrançois et al. 2020).

In this forum paper, we present the methodology of the workshops, and summarise the reflections developed and supported by the participants around the six main objectives that emerged from the discussions. This defines a strategy, or roadmap, for the successful implementation of DNA-based biomonitoring for freshwater ecosystems in Europe by 2030.

Method

The DNAqua-Net 2020 workshops were set up by a French team of five organisers under the lead of INRAE (Blancher P, Lefrançois E, Rimet F, Vasselon V and Bouchez A). The objectives of these workshops were to foresee the steps necessary for implementing DNA-based biomonitoring methods at European level and to foster the process of implementation. This was done through a three-fold approach: 1) identify actions to be taken in the different EU countries, 2) promote collaboration between countries, and 3) elaborate a shared strategy for the effective deployment of DNA-based methods at the European scale.

To this end, scientific experts, environmental authorities and policy-makers were brought together. At least one scientist and one representative of the competent authorities/agencies from the different countries involved in DNAqua-Net were invited. To enlarge this expert panel to include policy-makers, members of the "Water Framework Directive Common Implementation Strategy Working Group on Ecological Status" (ECOSTAT) were also invited. In the end, 50 participants representing 18 countries participated in the DNAqua-Net 2020 workshops (Fig. 1, Supp. material 1: Table S1). In order to ensure the best possible conditions for the active participation of all, eight online half-day web-workshops were organised in April 2020.

Each workshop had six to eight participants and two facilitators. After a general introduction and presentation of the participants, discussions were divided into three sections, each dealing with at least one of the six areas of action identified in the previous SYNAQUA workshops (Bouchez et al. 2021):

1. Development, experimentation and standardisation of DNA-based methods;
2. Regulation and organisation of bioassessment for successful implementation of eDNA-based methods;
3. Training and mobilisation of professionals;
4. Awareness-building and involvement of decision-makers and contracting authorities;
5. Awareness-building and involvement of the civil society;

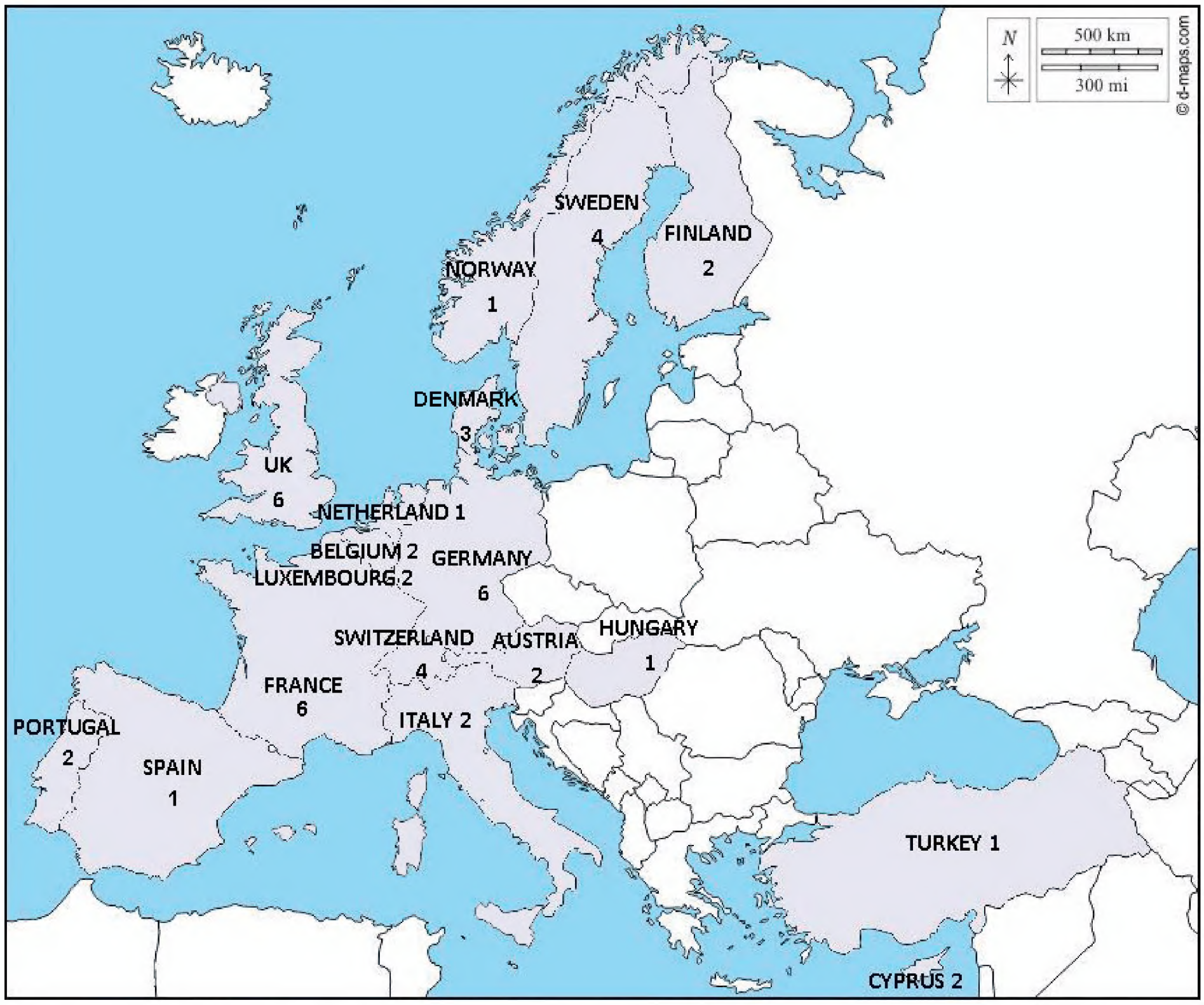

Figure 1. Countries of origin of the DNAqua-Net 2020 workshops participants and number of participants per country.

6. Need for a steering structure ensuring consistent and coordinated implementation.

Participants were asked to express their views: first, about their country's specific situation and concerns, and second, about needs at the EU level. Each workshop ended with a concluding session, and the minutes were shared with participants.

Results and discussions

The workshops distinguished between applications for regulatory purposes ('routine monitoring') and other applications of DNA-based methods in the context of restoration, conservation or research. For the first, i.e. routine monitoring, implementation must comply with the legislative requirements (e.g., of the EU Water Framework Directive (WFD)) and must demonstrate that it yields results that are in line with those produced by currently accepted methods (e.g., for WFD: Kelly et al. 2014; Poikane et al. 2014). However, at the same time, implementation should be able to adapt to harness new technical potential, if this does not conflict with the main principles. While the detailed points of discussion of all eight online workshops are documented in an open-access report (Lefrançois et al. 2020), we focus here on the key points that emerged. They provide the basis for a roadmap for implementing DNA-based methods for aquatic monitoring in Europe in relation to six main objectives (Fig. 2):

1. Highlight the effectiveness and benefits of DNA-based methods.
2. Develop an adaptive approach for successful implementation of new methods.
3. Provide best practice guidelines and standards.
4. Involve stakeholders and ensure good knowledge transfer.
5. Support the environmental biomonitoring sector to achieve the required changes.
6. Steer the process at European level.

No hierarchy among these objectives has been identified during the workshops; all should be implemented in parallel. The results of the workshop's discussions of these six objectives are outlined below.

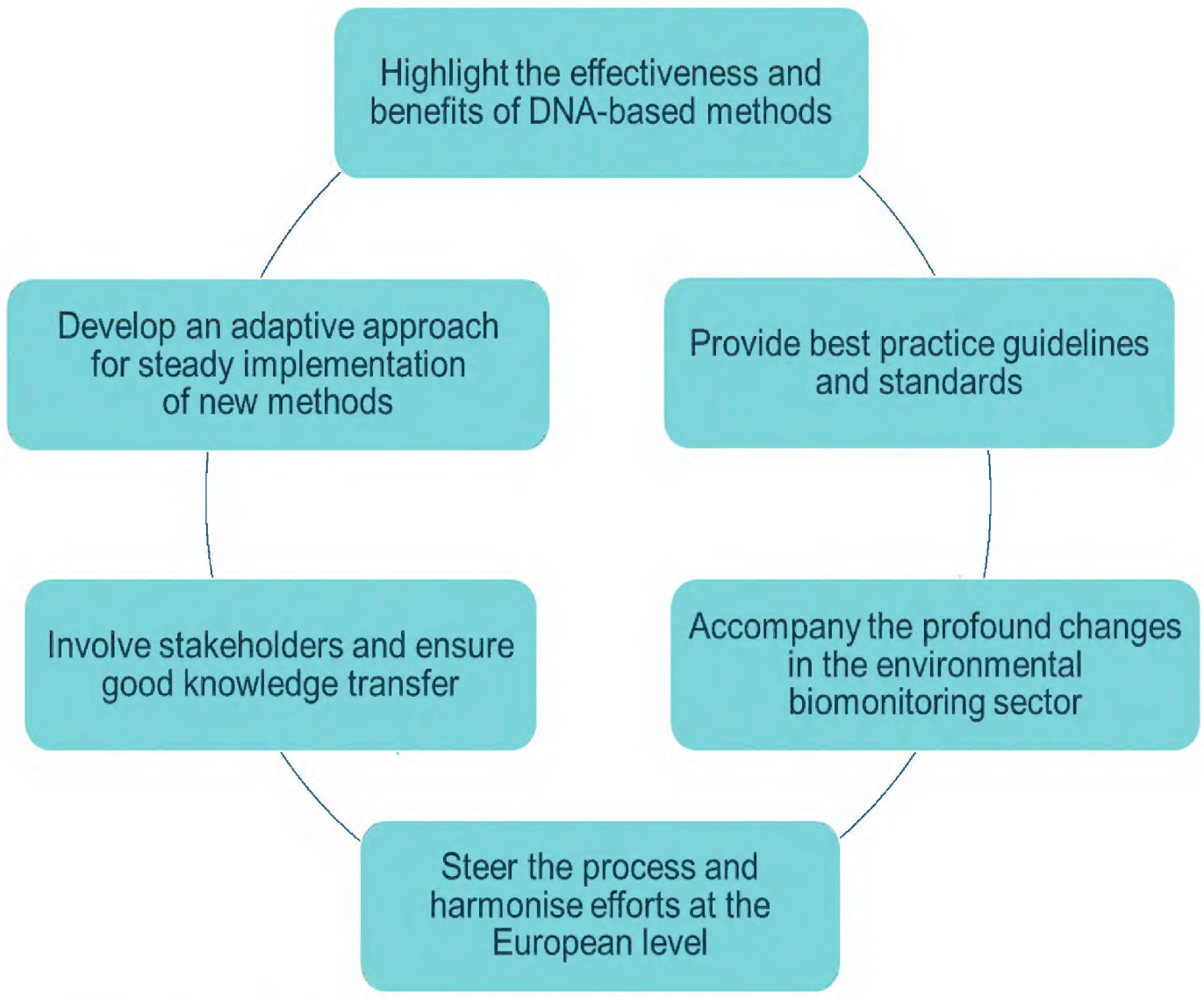
Highlight the effectiveness and benefits of DNA-based methods
Develop an adaptive approach for steady implementation of new methods
Provide best practice guidelines and standards
Involve stakeholders and ensure good knowledge transfer
Accompany the profound changes in the environmental biomonitoring sector
Steer the process and harmonise efforts at the European level

Figure 2. Components of an efficient implementation of DNA-based methods.

1. Highlight the effectiveness and benefits of DNA-based methods

DNA-based methods are now very popular, at least among researchers. However, workshop participants felt that, before their routine use for monitoring purposes, their effectiveness and added value compared to traditional methods, as well as their cost efficiency, still need to be accurately assessed and demonstrated, in particular for habitats and taxa where this has not yet been done rigorously. More generally, their benefits and limitations should be made clear to all stakeholders, taking into account their concerns (e.g., environmental goals, economic constraints). Indeed, for a relevant and effective demonstration, it is necessary to understand beforehand the real information needs of water managers and to try to meet them as best as possible. For instance, the non-invasive nature of some eDNA methods can be a strong argument. To clarify all these points, in Europe and beyond, numerous projects to compare traditional and DNA-based methods have been or are being implemented (e.g., Elbrecht et al. 2017; Pont et al. 2018; Bailet et al. 2019; Vasselon et al. 2019; Griffiths et al. 2021; Martins et al. 2021; Pissaridou et al. 2021; Weigand 2021).

The lack of confidence experienced in England following an abrupt change to a DNA-based method for the monitoring of phytobenthos (Kelly 2019) showed that the intercalibration of DNA-based methods with traditional methods must be organised over a wide spatial coverage but also over a sufficiently long period of overlap. However, molecular and morphological approaches assess biodiversity on different bases (Shaw et al. 2016; Vasselon et al. 2018; Apotheloz-Perret-Gentil et al. 2021), thus their intercalibration will not necessarily be clear and straightforward. Furthermore, there are almost as many traditional methods as there are countries (Birk et al. 2012), making the task even more complex. However, some studies are now showing that both approaches can detect broadly the same ecological gradients and their anthropogenic and environmental drivers (Tapolczai et al. 2018; Martins et al. 2021). These key results support the view that the two approaches can be meaningfully intercalibrated.

Thanks to numerous pilot projects throughout Europe (e.g., Mortágua et al. 2019; Pérez-Burillo et al. 2020; Meissner et al. 2021; Pissaridou et al. 2021), it is now commonly accepted that, from a general perspective and for the time being, traditional and DNA-based methods bring complementary information. However, when it comes to biodiversity estimates or detection of certain indicator taxa, in many cases the molecular method is often more accurate (e.g. Egeter et al. 2022).

Therefore, when traditional methods are already fulfilling the stakeholders' requirements, the implementation of DNA-based methods does not always appear as a priority for resource managers. On the other hand, for currently overlooked management issues, DNA approaches could have direct benefits such as:

- monitoring whole community impacts of restoration or management measures, monitoring ecosystem biodiversity, improving detection capabilities for invasive or endangered species to prompt quick management responses (e.g., Cantonati et al. 2020; Egeter et al. 2022);

- developing new metrics, able to measure the impacts of multiple pressures on the environment and to deliver a more accurate diagnosis of different human pressures, especially those less addressed (e.g., Poikane et al. 2020);
- providing biological assessment methods for habitats that currently lack tools (e.g., groundwater, transitional waters) or enabling large scale monitoring and screening of neglected waterbodies like small streams and small lakes (e.g., Bolpagni et al. 2019).

In addition, the implementation of DNA-based methods is also seen as an opportunity to reduce differences in ecosystem assessments between EU regions and countries, by offering new metrics where traditional methods have not been developed yet, where taxonomic expertise is lacking (e.g., Vasselon et al. 2017), or where harmonisation of methods is difficult. More globally, the development of new indices will be required to make full use of the wealth of information that molecular data can provide.

2. Develop an adaptive approach for successful implementation of new methods

Several DNA-based methods are now mature enough to be implemented in routine biomonitoring to meet end-user needs (Vasselon et al. 2019; Meissner et al. 2021). However, whatever the innovation, the transition phase from validation studies to full implementation is always delicate and needs to be well thought out.

Some participants preferred a "cautious approach" and recommended that implementation is halted until methods are fully tested, evaluated and calibrated. But optimisation of rapidly developing methods can be an endless task. Indeed, it is difficult to determine when a development can be considered sufficiently mature to move to the operational phase, even if some questions remain open.

Pragmatically, most participants in the workshops advocated an "adaptive approach", such as that being adopted in Switzerland (Pawlowski et al. 2020b), in which advances are relatively quickly implemented and tested in the field in an operational setting, adapted, and then validated for widespread use. At the same time, they stated the importance of comparing and harmonising methods if such a strategy is to succeed (Poikane et al. 2014).

Whatever strategy is adopted, "cautious" or "adaptive", there are two options for implementing the new methods (Hering et al. 2018):

- Like-for-like replacement of existing methods. In other words, use DNA-based methods as alternative means of data acquisition, but continuing with existing principles behind metrics, reference conditions and status class boundaries of WFD.
- Adoption of a "Biomonitoring 2.0" (Baird and Hajibabaei 2012), by developing a new generation of indices using the full potential of DNA data to explore the greater taxonomic depth that is available, use of a wider range of organisms, and potentially, developing metrics that measure ecosystem function as well as structure (Cordier et al. 2020).

While implementation of the first option would already require enormous effort (to achieve harmonisation), the second option is even more challenging (to preserve time series of environmental monitoring information, to create and calibrate new indices on accurate quality gradients starting from scratch, etc.). Indeed, the option described above as "new indices making use of the entire wealth of the molecular data", would open new avenues to improve the quality of ecological assessment (Apothéloz-Perret-Gentil et al. 2017; Pawlowski et al. 2018; Kelly et al. 2020) and ecosystem management.

Whatever strategy and option are adopted, it must primarily meet the needs of the stakeholders and be co-designed with them to decide "if" and "how" a new method should be implemented (see 4).

3. Provide best practice guidelines and standards

In groups of organisms and in countries where DNA-based methods are beginning to be used, problems related to the heterogeneity of practices have emerged and show the difficulty of comparing results. A need for best practice guidelines and standardisation at national and European levels was therefore identified by the DNAqua-Net community (Mergen et al. 2018). This need has begun to be addressed by the European Committee for Standardization (CEN), where two technical reports have already been published (e.g., CEN 2018). In 2019, CEN created a new working group dedicated to DNA and eDNA in TC 230 (WG28), which is currently consulting on a New Work Item Proposal for "*Sampling eDNA from Water Samples*" (CEN/TC 230 N 1229).

According to some participants, standardisation may raise several questions and also some fears, justified or not, such as:

- the difficulty of accommodating the organisational heterogeneity within particular nations or regions to suit the heterogeneity of national or even regional situations, as already experienced by some participants with traditional methods, notably in terms of data availability and format (e.g., format, controlled vocabularies);
- the risk of developing standards that are not suited to the operational context, in terms of technical and/or financial feasibility, and will therefore be ignored;
- the complexity and timeframe of such a standardisation process;
- the risk of hindering or preventing technological progress, because regulatory applications need some stability; biomonitoring may therefore not be able to benefit from the very rapid scientific and technological progress in this field.

All these problems are regularly solved in an industrial setting, but in the field of bioassessment, where it is important to trace changes in ecological status over time, it can be difficult to agree to allow standards to evolve with advances in technology.

These concerns should not prevent the necessary standardisation processes, but rather draw attention to how best practices should be developed and disseminated. Therefore, the participants recommended developing a flexible approach consisting of:

1. an overall framework based on guidelines for good methods and practices (Pawlowski et al. 2020b; Bruce et al. 2021), and interlaboratory ring tests (Vasselon et al. 2021);
2. specific methods and standards for key independent steps; e.g., diatom sampling (CEN 2018) or reference database (Rimet et al. 2021). This step-by-step approach is the one adopted by the CEN TC230/WG28.

Guidance documents, as well as training (see 4), should present the basic principles of eDNA analysis to all operators, so that they understand the constraints of biomolecular techniques and the resulting best practices to adopt. The EU should be a forerunner and a significant player in interlaboratory standardisation and methodological support through institutional (e.g., ECOSTAT, CEN) and scientific networks (e.g., DNAqua-Net). At national level, reference laboratories should be the guardians of quality and proficiency, responsible for training people, implementing interlaboratory tests, and testing new methods. Coordination of national efforts at the EU level would be beneficial (see 6). With the rapid development of methods, it is all the more important to intensify efforts in these directions.

4. Involve stakeholders and ensure good knowledge transfer

Conditions for the adoption of DNA-based methods vary among European countries and/or between taxonomic groups. Nevertheless, all European stakeholders need to gain confidence in DNA-based methods, as they still often see them as a black-box. Some understanding of bioinformatics pipelines and the temporal and spatial distributions of eDNA, and how these may affect interpretation, is especially important. To overcome such difficulties, transfer of knowledge and stakeholders' involvement is of great importance at all levels of governance, ranging from EU regulatory bodies to local authorities. This includes knowledge exchange between scientists, but also between scientists and stakeholders. For example, managers need to communicate their needs and outline their legal framework as well as their organisational and funding constraints. Scientists need to present a clear and understandable appraisal of methods, including their advantages and drawbacks. Thus, such exchanges may facilitate the outline of potential applications of scientific new methods, and on the other hand urge scientists to address stakeholders' specific needs.

To optimise knowledge transfer, research and development should focus on meeting the needs of the target audiences. These may differ among stakeholders and may evolve over time, and must be well understood by scientists. It also needs to be taken into account that stakeholders are sensitive not only to the efficiency of different methods, but also to a combination of contextual parameters that scientists often perceive poorly: costs, public policies, political context, risk of EU litigation, lobbying pressures, etc.

The gap between the development of methods by scientists and their use by managers is an issue that has been raised in many fields (e.g., water, soil, landscape, etc.). Often, the methods developed by scientists are not used by managers, and this is not only a question of knowledge transfer. It is actually because the general approach to the development of these methods needs to be rethought in order to be more strategic and systematic (Slunge et al. 2017) and more inclusive (Schmidt et al. 2020). Indeed, given the numerous actors and diverse expertise involved, the implementation of DNA-based methods in bioassessment is a major transdisciplinary effort that would benefit enormously from early interactions undertaken in a spirit of cooperation and creativity (Slunge et al. 2017). Several studies have shown that the knowledge and experience of stakeholders and scientists are complementary, and that both are needed to co-create methods for assessing environmental quality (e.g., Reyjol et al. 2014). This will make it possible, on the one hand, to adapt methods to the needs thus identified and, on the other hand, to effectively inform stakeholders about existing methods. Development must also go hand-in-hand with implementation through, for example, joint planning, pilot studies, demonstrations, experiments, joint training sessions between the various stakeholders, and steering committees, etc. Such a culture of genuine co-creation must be developed so that informed choices about bioindication and biomonitoring are made at all levels.

In co-creative settings, communication and training also have to be effective. Hence, training courses have to meet different objectives depending on the target audience, e.g.:

- *Deliver basic knowledge to all stakeholders* to enable them to discuss the topic and have dialogue with other actors: what exactly it is, scientific interest, applications, benefits and limitations, etc.
- *Provide more in-depth and accurate knowledge to the particular stakeholders who oversee and manage* the deployment of these methods (such as administrations, standardisation bodies, etc.). They need to gain a clear understanding of the added value of DNA-based methods, helping them to identify the specific situations (taxa, habitats) where i) the two approaches are complementary (even if the overall costs are increased), ii) they are interchangeable, iii) one is better than the other. They also need to develop the capacity to check the quality of providers and the traceability of results. Information on the repeatability, limits, uncertainty and control points of the methods are required.

- *Ensure full control of these techniques to operators* that implement them. These key-actors implement the sampling in the field, the sample analysis in the lab and the sample interpretation (bioinformatics and ecological interpretation). They need expert training to fully control these steps, in order to produce accurate and informative results.

Several participants noted that it is very challenging to get a comprehensive knowledge of what is done at the European scale in the field of eDNA application. They thought it might be of interest to list all the DNA-based methods that are currently available or developed for each biological compartment and each waterbody, and make them available to a larger audience. Also, a catalogue of competent research laboratories, service providers, as well as existing guides and normative documents is needed. A first attempt to meet this need has been made with the launch of the DNAqua-Hub platform (https://dnaquahub.eu/). Such platforms should be regularly and collaboratively updated and enriched to follow new developments, methods' implementations, and the new environmental biomonitoring market.

5. Support the environmental biomonitoring sector as it adjusts to the changes required

The implementation of DNA-based methods certainly calls for a significant reorganisation of the bioassessment sector, depending on how it is currently organised in each country. In some countries, most services are outsourced to the private sector (e.g., Finland, France, Cyprus). In that case, a widely shared observation is that it is a big risk today for small to medium-sized private companies to invest and get involved in rapidly evolving approaches like DNA-based biomonitoring. Favourable conditions for such investments need to be created through both regulatory demands, method standardisation and starter funding. Elsewhere, changes have historically been more likely to be supported by administrations (e.g., England). Whatever the context, funding is a key issue, due to the current economic strategy of many countries to reduce their investment in bioassessment. There is a need to demonstrate that the implementation of DNA-based methods is not only a cost-effective solution, but that it can be used to improve biomonitoring and provide additional, useful information for decision makers, while also addressing citizens' concerns, such as climate and environmental changes.

Among possible consequences of changes in the biomonitoring sector, the evolution towards molecular methods may reduce the number of experts needed to identify routine samples in administrations or operators. Therefore, there is a risk that the organisations concerned, will no longer retain or appoint people with expertise in taxonomy and ecology, whose skills could be wrongly considered as no longer useful. The forum emphasised that, whatever the method used to obtain biomonitoring data (morphology or DNA-based), it is essential that these data are properly interpreted by people with expertise in aquatic ecology. Taxonomy and ecology expertise is essential to interpret the data from molecular studies, and to enable intercalibrations. In the future, experts will still be needed in these fields (e.g. national focus taxonomic experts to regularly control that the methods are holding good standards, and to be able to interpret the results). This will contribute to the need expressed by participants to develop a "bioassessment culture" leading to optimal biomonitoring and ecosystem management, a culture that is goal-oriented, aiming to understand environmental changes and proposing targeted measures that take into account both cost-benefit analysis and a concern for practicality.

In practice, the national level seems to be the appropriate one to integrate these new methods into monitoring programmes. However, upstream agreement by the EU on general monitoring method requirements is vital. This should preferably be done through the establishment of overall CEN standards, which can then be mirrored in each EU country. This will avoid unnecessary methodological pluralism and provide stability to the market, which is essential to attract necessary private sector investments. For regulatory monitoring under the WFD, any change of method requires intercalibration, which involves significant procedures (e.g., carefully designed programmes in which matching samples are analysed in parallel using different protocols) and effort. This makes it difficult to change methods within a management cycle, i.e., before 2027 for the current cycle. Even then, as member states are at different stages of biomonitoring sophistication, national uptake of new molecular methods into national monitoring programs will proceed gradually and with varying speeds. Broad European cooperation will be key to enable a switch to molecular monitoring methods beyond the current management cycle. This cooperation will be particularly needed in the fields of methods' validation, DNA-based data management and related standardisation.

6. Steer the process at European level

All workshop participants considered that we are now at a stage where the development of some DNA-based biomonitoring methods is well advanced and is attracting interest from stakeholders in many countries. Therefore, they considered that now is a good time to create a steering group at European level. For example, a sub-group of experts on DNA-based methods in ECOSTAT could guide future changes in the assessment of the ecological status of waterbodies in Europe. This would offer an opportunity to discuss and advance the implementation of DNA-based methods, to provide guidance, and to encourage Member States to develop DNA-based methods for ecological status assessment where appropriate.

Such a European steering group should have a clear mandate and contribute, for example, to:

- share knowledge, produce recommendations, explain what can be done with DNA-based methods, how the implementation of the WFD can be improved, etc.;
- develop a strategy to compare and harmonise DNA-based methods, i.e., to intercalibrate them to ensure common management objectives across Europe;

- produce a realistic implementation scenario;
- provide a common framework for Member States to develop their own (intercalibrated) indices and national monitoring strategy.

Working together at the European level would allow costs and benefits and experiences to be shared across borders, avoiding unnecessary overlap and effort. It could also help secure and raise national and European funding for applied research projects that involve and promote better transnational interaction, e.g., sharing samples and methods. A recent example was the 4th Joint Danube Survey, with the application of DNA-based approaches in addition to traditional morphology-based ones (Weigand 2021).

Conclusion

In its Environmental Implementation Review (European Commission 2019), the EU calls on Member States to step up efforts to improve water quality. According to the most recent statistics (EEA 2018), only about 40% of surface waters in Europe have achieved good or high ecological status as required by the WFD. Faced with the loss of biodiversity, the degradation of aquatic ecosystems and the services they provide, amplified by the impacts of climate change, actions must be taken (IPBES 2019). High-performance diagnostic and monitoring tools are needed to provide an effective scorecard and ensure the efficiency of preservation and restoration programmes.

To take full advantage of DNA-based methods, the DNAqua-Net workshops' participants call for urgent and purposeful implementation of a strategy built around the six complementary goals they identify (Fig. 2). In order to contribute to this strategy, it would be valuable to extend the work undertaken by DNAqua-Net, produce step-by-step guidelines and transmit them to practitioners, and show and communicate the different approaches, their advantages and benefits, etc. This could be facilitated via new EU research and network funding in support of the Green Deal and Biodiversity strategy for 2030.

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Supplementary material 1

Table S1

Author: Philippe Blancher, Estelle Lefrançois, Frédéric Rimet, Valentin Vasselon, Christine Argillier, Jens Arle, Pedro Beja, Pieter Boets, Jeanne Boughaba, Christian Chauvin, Michael Deacon, Willie Duncan, Gunilla Ejdung, Stefania Erba, Benoit Ferrari, Helmut Fischer, Bernd Hänfling, Michael Haldin, Daniel Hering, Nicolas Hette-Tronquart, Alice Hiley, Marko Järvinen, Benjamin Jeannot, Maria Kahlert, Martyn Kelly, Julia Kleinteich, Serdar Koyuncuoğlu, Sascha Krenek, Sidsel Langhein-Winther, Florian Leese, David Mann, Rémy Marcel, Stefania Marcheggiani, Kristian Meissner, Patricia Mergen, Olivier Monnier, Frank Narendja, Diane Neu, Veronica Onofre Pinto, Alina Pawlowska, Jan Pawlowski, Martin Petersen, Sandra Poikane, Didier Pont, Marie-Sophie Renevier, Steinar Sandoy, Jonas Svensson, Rosa Trobajo, Andrea Tünde Zagyva, Iakovos Tziortzis, Berry van der Hoorn, Marlen Ines Vasquez, Kerry Walsh, Alexander Weigand, Agnès Bouchez

Data type: pdf file

Explanation note: DNAqua-Net 2020 workshops participants (* Current or former member of the European Ecological Status Working Group of experts (ECOSTAT), that deals with issues regarding the ecological status for the implementation of Water Framework Directive, as a part of the Common Implementation Strategy).

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